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AN OUTBREAK OF ROUP AND CHICKEN-POX IN WHICH THE HIGH MORTALITY WAS APPAR- ENTLY CAUSED BY A SECONDARY INVADER *

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Some outbreaks of roup and chicken-pox are attended by a high mortality; at times 85-90% of the affected birds succumb. In other outbreaks the mortality may be only 1-2%. Observations made in a number of outbreaks in which the mortality varied greatly, revealed no essential difference either as to character or extent in the lesions.

Death supposedly results from an absorption of toxins or poisons from the affected areas. This, in many instances, is perhaps the case, especially in those outbreaks attended by a low mortality, as recovery seems to be dependent largely on the extent of the lesions. (The general health of some individuals remains good in spite of extensive lesions. These cases usually recover. With this exception, the general statement holds good.) It appears hardly possible, however, that such is the case where the mortality runs high, as many birds die very suddenly before the development of extensive lesions.

An investigation by the authors of an outbreak which occurred in a large flock of poultry apparently throws some light on the subject, at least as far as this one outbreak is concerned. Following is a digest of the reports of the poultryman in charge of the plant at the time the outbreak occurred.

He purchased a number of hens which became sick shortly afterward with roup and chicken-pox. He noticed that they showed canker. A few days later other hens became sick, some showing canker, some pox scabs, and some both. Shortly after the appearance of these lesions, the hens began to die. The mortality ran at times as high as 30 to 120 per week. The poultryman was unable, as far as he could tell, to save any that showed canker. Nearly 2,000 chickens died in less than 10 months.

A bacteriologic examination of material from these lesions revealed so many different types of organisms that nothing of note was determined. However, a hen that was injected subcutaneously with a suspension obtained by macerat-

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ing in normal salt solution material from the canker lesions and pox scabs, became sick in 24 hours, moved very little, would not eat, developed a diarrhea on the second day, and in about 72 hours died. It was thought at this time that death was probably due to septic infection. The postmortem findings in this case were slight emaciation; catarrhal exudate in the buccal cavity and about the external nares; visceral peritoneum and oviduct slightly congested; abdominal organs otherwise normal; the right heart normal, the left dilated and filled with a semifluid dark blood clot; lungs pink in color and inflated.

Stained smears from the heart's blood, when examined under the microscope, revealed a cocco-bacillus 1 micron long by 0.5-1 micron wide. It would stain by the ordinary aniline dyes. Morphologically the organism resembled that of chicken-cholera. A marked leukocytosis was noted. Cultures from the heart's blood and liver on neutral agar revealed, after 24 hours, a very thin, scanty growth, translucent, and slightly beaded in appearance. Slightly heavier growth was obtained on 2% raw serum agar, a very scant growth on glycerin and lactose agar and dextrose broth, while plain broth and plain and litmus milk revealed no growth. Stained preparations from artificial media under the microscope appeared identical with those stained from the blood.

Subcultures could not be obtained except on serum agar, and then only a scanty growth. With a few exceptions, the cultures would die in from 2 to 6 days after isolation, and rarely would subcultures show any growth, even on serum agar. In order to propagate this organism, it was necessary to inject a chicken about every fourth day. Hen C1000 was injected, April 9, with a culture isolated 4 days previously, but remained well. On April 13, this same hen was injected with a culture isolated the day before. Six days later she died and the organism was recovered from the blood.

In some respects this bacillus resembles that of chicken-cholera, in that a bipolar staining is at times seen in preparations direct from the blood and it produces an acute septicemia in chickens, but differs from it markedly in its cultural characteristics in the fact that ducks are immune and injections of the killed culture will render a hen immune to subsequent injections of the living culture, which, however, will not be immune to chicken-cholera.

This organism does not cause roup or chicken-pox.—In no instance were any lesions of roup or chicken-pox produced by infection.

Injection of a killed culture confers immunity.—Hen C987, given 6 c.c. of a heated suspension, 16 days later withstood an inoculation with a virulent culture to which Hen C1000, injected at the same time, succumbed. And again, Hen A100, given 3 c.c. of the heated suspension, 7 days later withstood an inoculation of the living organism, which killed Hen A22, injected simultaneously.

Ducks are immune.—A mallard duck injected with Culture A373 remained well, while at the same time a pigeon succumbed to the

infection. Another mallard duck withstood an injection that killed Hen A316. Indian Runner Duck 983 withstood the injection that killed Hen 486.

Ducks immune to this organism succumb to fowl-cholera.—Ducks 1 and 983, as noted, withstood injections (subcutaneous) of this organism, but later succumbed to fowl-cholera.

This organism will not confer immunity to fowl-cholera.—It will be noted in the accompanying table that altho Hen C987, injected with a large quantity of killed culture, was subsequently immune to the living culture which killed Hen C989 injected at the same time, nevertheless Hen C987 when injected with fowl-cholera promptly died.

Hen	Culture Injected	Result
C987	6 c.c. of heated suspension* from infected heads	Remained well
C987	Culture A284	Remained well
C987	1 c.c. suspension of fowl- cholera from Rhode Island	Died 3 days later
C989	1.5 c.c. of unheated culture from Vermont hens	Died 4 days later. Organism isolated from heart's blood

* Injected subcutaneously.

Animals susceptible.—Besides chickens and ducks already mentioned, rabbits, mice, guinea-pigs, and pigeons were injected as follows:

Animal	Culture Injected	Days Intervening Before Death	Source of Cultures Obtained
Rabbit	A284†	1	—
Two mice	A284	(1st) 1; (2nd) 2	—
Guinea-pig	Culture isolated the day before injection	7	—
Rabbit	Culture of the day before injection	1	Heart
Pigeon	A373	2	Heart
Pigeon	Culture of the day before injection	2	Heart

† Suspension so light that no turbidity was seen.

Portals of entry.—Our experiments show that this disease is one of wound infection. It has been produced 14 times by subcutaneous injections in chickens alone as shown by the accompanying table:

Hen	Culture Injected	Days Intervening Before Death	Source of Cultures Obtained
A360	2 c.c. of light suspension of Culture C989	3	Heart and liver
A287	C989	2	Heart
A284	Culture isolated 4 days before injection	2	Heart's blood
A22	1.5 c.c. A287	Sick, but finally recovered	
C1000	Culture A284 isolated the day before injection	6	Organism found in blood
A31	Culture of the day before injection	2	Heart
A373	Culture of the day before injection	2	
A254	Mixed culture	2	Heart
A316	Mixed culture	4	Heart
A49	174 and A316	1	Heart's blood
486	174 and A49	2	Heart
A99	Mixed culture	9	Heart
200	Blood tube from Hen A316	1	Heart
Ckl. C1321	Blood tube of Hen A99	6	Cultures made

An old scab was removed from the comb of Hen 174 and some virulent culture instilled. Death occurred 5 days later. A fresh wound was made on the comb and wattles of Hen A305 and some of the virulent culture instilled. This hen, however, remained well, as a result possibly of the blood's mechanically washing the organism from the wound.

The organism will not penetrate the unabraded mucous membrane when instilled into the eye and nostrils.

Hen	Seat of Inoculation	Culture	Result
A22	Eye	C989	Remained well
A246	Nostril	C989	Remained well

We were unable to produce the disease by feeding either the virulent culture or fecal material from affected birds.

Hen	Culture	Result
A294	A373	Remained well
A33	A316	Remained well
A273	A49	Remained well
104	Fed fecal matter from Hen 174	Remained well

The data which we could gather as regards the biology of this organism, indicate that it probably belongs to the hemorrhagic septiemia group. As pointed out, under the microscope it resembles fowl-cholera but differs from it markedly in two respects:

1. Cultural characteristics. Growth was very meager, while fowl-cholera exhibits much heavier growth, will live much longer outside the animal body, and has a different appearance culturally when grown on agar slopes.

2. Pathogenicity. As already pointed out, ducks are immune to this organism, and the injection of killed cultures confers no immunity to fowl-cholera. This is a disease of wound infection, while fowl-cholera may be transmitted by way of the mouth.

Just what relation secondary invaders have to roup and chicken-pox in general throughout the country is hard to state, as unfortunately we have not had opportunity to investigate another outbreak where the mortality ran high. However, it seems reasonable to suppose that their rôle is far from inconsequential when we consider the wide range in mortality in different outbreaks in which the lesions both as to character and extent are similar.